

# SEQUENCE LISTING

<110> Wang, Yan  
 Xi, Lei  
 Prosen, Dennis E.  
 MJ Bioworks, Inc.

<120> Improved Nucleic Acid Modifying Enzymes

<130> 020130-000111US

<140> US 09/870,353

<141> 2001-05-30

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<151> 2000-05-26

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<170> PatentIn Ver. 2.1

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 binding protein Ssod7d

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<210> 8  
 <211> 844  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Description of Artificial Sequence:fusion protein  
 Pfu-Ssod7d

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Thr Phe Arg Pro Tyr Ile Tyr Ala Leu Leu Arg Asp Asp Ser Lys Ile
             35             40             45
Glu Glu Val Lys Lys Ile Thr Gly Glu Arg His Gly Lys Ile Val Arg
             50             55             60
Ile Val Asp Val Glu Lys Val Glu Lys Lys Phe Leu Gly Lys Pro Ile
             65             70             75             80
Thr Val Trp Lys Leu Tyr Leu Glu His Pro Gln Asp Val Pro Thr Ile
             85             90             95
Arg Glu Lys Val Arg Glu His Pro Ala Val Val Asp Ile Phe Glu Tyr
             100            105            110
Asp Ile Pro Phe Ala Lys Arg Tyr Leu Ile Asp Lys Gly Leu Ile Pro
             115            120            125

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Met	Glu	Gly	Glu	Glu	Glu	Leu	Lys	Ile	Leu	Ala	Phe	Asp	Ile	Glu	Thr	130	135	140
Leu	Tyr	His	Glu	Gly	Glu	Glu	Phe	Gly	Lys	Gly	Pro	Ile	Ile	Met	Ile	145	150	155
Ser	Tyr	Ala	Asp	Glu	Asn	Glu	Ala	Lys	Val	Ile	Thr	Trp	Lys	Asn	Ile	165	170	175
Asp	Leu	Pro	Tyr	Val	Glu	Val	Val	Ser	Ser	Glu	Arg	Glu	Met	Ile	Lys	180	185	190
Arg	Phe	Leu	Arg	Ile	Ile	Arg	Glu	Lys	Asp	Pro	Asp	Ile	Ile	Val	Thr	195	200	205
Tyr	Asn	Gly	Asp	Ser	Phe	Asp	Phe	Pro	Tyr	Leu	Ala	Lys	Arg	Ala	Glu	210	215	220
Lys	Leu	Gly	Ile	Lys	Leu	Thr	Ile	Gly	Arg	Asp	Gly	Ser	Glu	Pro	Lys	225	230	235
Met	Gln	Arg	Ile	Gly	Asp	Met	Thr	Ala	Val	Glu	Val	Lys	Gly	Arg	Ile	245	250	255
His	Phe	Asp	Leu	Tyr	His	Val	Ile	Thr	Arg	Thr	Ile	Asn	Leu	Pro	Thr	260	265	270
Tyr	Thr	Leu	Glu	Ala	Val	Tyr	Glu	Ala	Ile	Phe	Gly	Lys	Pro	Lys	Glu	275	280	285
Lys	Val	Tyr	Ala	Asp	Glu	Ile	Ala	Lys	Ala	Trp	Glu	Ser	Gly	Glu	Asn	290	295	300
Leu	Glu	Arg	Val	Ala	Lys	Tyr	Ser	Met	Glu	Asp	Ala	Lys	Ala	Thr	Tyr	305	310	315
Glu	Leu	Gly	Lys	Glu	Phe	Leu	Pro	Met	Glu	Ile	Gln	Leu	Ser	Arg	Leu	325	330	335
Val	Gly	Gln	Pro	Leu	Trp	Asp	Val	Ser	Arg	Ser	Ser	Thr	Gly	Asn	Leu	340	345	350
Val	Glu	Trp	Phe	Leu	Leu	Arg	Lys	Ala	Tyr	Glu	Arg	Asn	Glu	Val	Ala	355	360	365
Pro	Asn	Lys	Pro	Ser	Glu	Glu	Glu	Tyr	Gln	Arg	Arg	Leu	Arg	Glu	Ser	370	375	380
Tyr	Thr	Gly	Gly	Phe	Val	Lys	Glu	Pro	Glu	Lys	Gly	Leu	Trp	Glu	Asn	385	390	395
Ile	Val	Tyr	Leu	Asp	Phe	Arg	Ala	Leu	Tyr	Pro	Ser	Ile	Ile	Ile	Thr	405	410	415
His	Asn	Val	Ser	Pro	Asp	Thr	Leu	Asn	Leu	Glu	Gly	Cys	Lys	Asn	Tyr	420	425	430
Asp	Ile	Ala	Pro	Gln	Val	Gly	His	Lys	Phe	Cys	Lys	Asp	Ile	Pro	Gly	435	440	445
Phe	Ile	Pro	Ser	Leu	Leu	Gly	His	Leu	Leu	Glu	Glu	Arg	Gln	Lys	Ile	450	455	460
Lys	Thr	Lys	Met	Lys	Glu	Thr	Gln	Asp	Pro	Ile	Glu	Lys	Ile	Leu	Leu	465	470	475
Asp	Tyr	Arg	Gln	Lys	Ala	Ile	Lys	Leu	Leu	Ala	Asn	Ser	Phe	Tyr	Gly	485	490	495
Tyr	Tyr	Gly	Tyr	Ala	Lys	Ala	Arg	Trp	Tyr	Cys	Lys	Glu	Cys	Ala	Glu	500	505	510
Ser	Val	Thr	Ala	Trp	Gly	Arg	Lys	Tyr	Ile	Glu	Leu	Val	Trp	Lys	Glu	515	520	525
Leu	Glu	Glu	Lys	Phe	Gly	Phe	Lys	Val	Leu	Tyr	Ile	Asp	Thr	Asp	Gly	530	535	540
Leu	Tyr	Ala	Thr	Ile	Pro	Gly	Gly	Glu	Ser	Glu	Glu	Ile	Lys	Lys	Lys	545	550	555
Ala	Leu	Glu	Phe	Val	Lys	Tyr	Ile	Asn	Ser	Lys	Leu	Pro	Gly	Leu	Leu	565	570	575
Glu	Leu	Glu	Tyr	Glu	Gly	Phe	Tyr	Lys	Arg	Gly	Phe	Phe	Val	Thr	Lys	580	585	590
Lys	Arg	Tyr	Ala	Val	Ile	Asp	Glu	Gly	Lys	Val	Ile	Thr	Arg	Gly		595	600	605



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ccgtgcccct ggaggtggag gtggggatag gggaggactg gctctccgcc aaggagggca 1860
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<210> 10

<211> 634

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: fusion protein  
Sac7d-deltaTaq

<400> 10

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Glu Lys Glu Val Asp Thr Ser Lys Ile Lys Lys Val Trp Arg Val Gly
          20           25           30
Lys Met Val Ser Phe Thr Tyr Asp Asn Gly Lys Thr Gly Arg Gly
          35           40           45
Ala Val Ser Glu Lys Asp Ala Pro Lys Glu Leu Leu Asp Met Leu Ala
          50           55           60
Arg Ala Glu Arg Glu Lys Lys Gly Gly Gly Val Thr Ser Pro Lys Ala
          65           70           75           80
Leu Glu Glu Ala Pro Trp Pro Pro Pro Glu Gly Ala Phe Val Gly Phe
          85           90           95
Val Leu Ser Arg Lys Glu Pro Met Trp Ala Asp Leu Leu Ala Leu Ala
          100          105          110
Ala Ala Arg Gly Gly Arg Val His Arg Ala Pro Glu Pro Tyr Lys Ala
          115          120          125
Leu Arg Asp Leu Lys Glu Ala Arg Gly Leu Leu Ala Lys Asp Leu Ser
          130          135          140
Val Leu Ala Leu Arg Glu Gly Leu Gly Leu Pro Pro Gly Asp Asp Pro
          145          150          155          160
Met Leu Leu Ala Tyr Leu Leu Asp Pro Ser Asn Thr Thr Pro Glu Gly
          165          170          175
Val Ala Arg Arg Tyr Gly Gly Glu Trp Thr Glu Glu Ala Gly Glu Arg
          180          185          190
Ala Ala Leu Ser Glu Arg Leu Phe Ala Asn Leu Trp Gly Arg Leu Glu
          195          200          205
Gly Glu Glu Arg Leu Leu Trp Leu Tyr Arg Glu Val Glu Arg Pro Leu
          210          215          220
Ser Ala Val Leu Ala His Met Glu Ala Thr Gly Val Arg Leu Asp Val
          225          230          235          240

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Ala Tyr Leu Arg Ala Leu Ser Leu Glu Val Ala Glu Glu Ile Ala Arg  
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 Leu Glu Ala Glu Val Phe Arg Leu Ala Gly His Pro Phe Asn Leu Asn  
 260 265 270  
 Ser Arg Asp Gln Leu Glu Arg Val Leu Phe Asp Glu Leu Gly Leu Pro  
 275 280 285  
 Ala Ile Gly Lys Thr Glu Lys Thr Gly Lys Arg Ser Thr Ser Ala Ala  
 290 295 300  
 Val Leu Glu Ala Leu Arg Glu Ala His Pro Ile Val Glu Lys Ile Leu  
 305 310 315 320  
 Gln Tyr Arg Glu Leu Thr Lys Leu Lys Ser Thr Tyr Ile Asp Pro Leu  
 325 330 335  
 Pro Asp Leu Ile His Pro Arg Thr Gly Arg Leu His Thr Arg Phe Asn  
 340 345 350  
 Gln Thr Ala Thr Ala Thr Gly Arg Leu Ser Ser Ser Asp Pro Asn Leu  
 355 360 365  
 Gln Asn Ile Pro Val Arg Thr Pro Leu Gly Gln Arg Ile Arg Arg Ala  
 370 375 380  
 Phe Ile Ala Glu Glu Gly Trp Leu Leu Val Ala Leu Asp Tyr Ser Gln  
 385 390 395 400  
 Ile Glu Leu Arg Val Leu Ala His Leu Ser Gly Asp Glu Asn Leu Ile  
 405 410 415  
 Arg Val Phe Gln Glu Gly Arg Asp Ile His Thr Glu Thr Ala Ser Trp  
 420 425 430  
 Met Phe Gly Val Pro Arg Glu Ala Val Asp Pro Leu Met Arg Arg Ala  
 435 440 445  
 Ala Lys Thr Ile Asn Phe Gly Val Leu Tyr Gly Met Ser Ala His Arg  
 450 455 460  
 Leu Ser Gln Glu Leu Ala Ile Pro Tyr Glu Glu Ala Gln Ala Phe Ile  
 465 470 475 480  
 Glu Arg Tyr Phe Gln Ser Phe Pro Lys Val Arg Ala Trp Ile Glu Lys  
 485 490 495  
 Thr Leu Glu Glu Gly Arg Arg Arg Gly Tyr Val Glu Thr Leu Phe Gly  
 500 505 510  
 Arg Arg Arg Tyr Val Pro Asp Leu Glu Ala Arg Val Lys Ser Val Arg  
 515 520 525  
 Glu Ala Ala Glu Arg Met Ala Phe Asn Met Pro Val Gln Gly Thr Ala  
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 Ala Asp Leu Met Lys Leu Ala Met Val Lys Leu Phe Pro Arg Leu Glu  
 545 550 555 560  
 Glu Met Gly Ala Arg Met Leu Leu Gln Val His Asp Glu Leu Val Leu  
 565 570 575  
 Glu Ala Pro Lys Glu Arg Ala Glu Ala Val Ala Arg Leu Ala Lys Glu  
 580 585 590  
 Val Met Glu Gly Val Tyr Pro Leu Ala Val Pro Leu Glu Val Glu Val  
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<210> 11

<211> 1965

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: fusion protein  
 PL-deltaTaq

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 gaggtagaca tctccaagat caagaaagta tggcgtgtgg gcaagatgat ctcccttcacc 180  
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 gccgtgcccc tggaggtgga ggtggggata ggggaggact ggctctccgc caaggagggc 1920  
 attgatggcc gcggcgaggc cgggcatcat catcatcctc attaa 1965

<210> 12  
 <211> 654  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Description of Artificial Sequence:fusion protein  
 PL-deltaTaq

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 20 25 30  
 Phe Lys Tyr Lys Gly Glu Glu Lys Glu Val Asp Ile Ser Lys Ile Lys  
 35 40 45  
 Lys Val Trp Arg Val Gly Lys Met Ile Ser Phe Thr Tyr Asp Glu Gly  
 50 55 60  
 Gly Gly Lys Thr Gly Arg Gly Ala Val Ser Glu Lys Asp Ala Pro Lys  
 65 70 75 80

Glu	Leu	Leu	Gln	Met	Leu	Glu	Lys	Gln	Lys	Lys	Gly	Gly	Gly	Val	Thr	85	90	95
Ser	Pro	Lys	Ala	Leu	Glu	Glu	Ala	Pro	Trp	Pro	Pro	Pro	Glu	Gly	Ala	100	105	110
Phe	Val	Gly	Phe	Val	Leu	Ser	Arg	Lys	Glu	Pro	Met	Trp	Ala	Asp	Leu	115	120	125
Leu	Ala	Leu	Ala	Ala	Ala	Arg	Gly	Gly	Arg	Val	His	Arg	Ala	Pro	Glu	130	135	140
Pro	Tyr	Lys	Ala	Leu	Arg	Asp	Leu	Lys	Glu	Ala	Arg	Gly	Leu	Leu	Ala	145	150	155
Lys	Asp	Leu	Ser	Val	Leu	Ala	Leu	Arg	Glu	Gly	Leu	Gly	Leu	Pro	Pro	165	170	175
Gly	Asp	Asp	Pro	Met	Leu	Leu	Ala	Tyr	Leu	Leu	Asp	Pro	Ser	Asn	Thr	180	185	190
Thr	Pro	Glu	Gly	Val	Ala	Arg	Arg	Tyr	Gly	Gly	Glu	Trp	Thr	Glu	Glu	195	200	205
Ala	Gly	Glu	Arg	Ala	Ala	Leu	Ser	Glu	Arg	Leu	Phe	Ala	Asn	Leu	Trp	210	215	220
Gly	Arg	Leu	Glu	Gly	Glu	Glu	Arg	Leu	Leu	Trp	Leu	Tyr	Arg	Glu	Val	225	230	235
Glu	Arg	Pro	Leu	Ser	Ala	Val	Leu	Ala	His	Met	Glu	Ala	Thr	Gly	Val	245	250	255
Arg	Leu	Asp	Val	Ala	Tyr	Leu	Arg	Ala	Leu	Ser	Leu	Glu	Val	Ala	Glu	260	265	270
Glu	Ile	Ala	Arg	Leu	Glu	Ala	Glu	Val	Phe	Arg	Leu	Ala	Gly	His	Pro	275	280	285
Phe	Asn	Leu	Asn	Ser	Arg	Asp	Gln	Leu	Glu	Arg	Val	Leu	Phe	Asp	Glu	290	295	300
Leu	Gly	Leu	Pro	Ala	Ile	Gly	Lys	Thr	Glu	Lys	Thr	Gly	Lys	Arg	Ser	305	310	315
Thr	Ser	Ala	Ala	Val	Leu	Glu	Ala	Leu	Arg	Glu	Ala	His	Pro	Ile	Val	325	330	335
Glu	Lys	Ile	Leu	Gln	Tyr	Arg	Glu	Leu	Thr	Lys	Leu	Lys	Ser	Thr	Tyr	340	345	350
Ile	Asp	Pro	Leu	Pro	Asp	Leu	Ile	His	Pro	Arg	Thr	Gly	Arg	Leu	His	355	360	365
Thr	Arg	Phe	Asn	Gln	Thr	Ala	Thr	Ala	Thr	Gly	Arg	Leu	Ser	Ser	Ser	370	375	380
Asp	Pro	Asn	Leu	Gln	Asn	Ile	Pro	Val	Arg	Thr	Pro	Leu	Gly	Gln	Arg	385	390	395
Ile	Arg	Arg	Ala	Phe	Ile	Ala	Glu	Glu	Gly	Trp	Leu	Leu	Val	Ala	Leu	405	410	415
Asp	Tyr	Ser	Gln	Ile	Glu	Leu	Arg	Val	Leu	Ala	His	Leu	Ser	Gly	Asp	420	425	430
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Thr	Ala	Ser	Trp	Met	Phe	Gly	Val	Pro	Arg	Glu	Ala	Val	Asp	Pro	Leu	450	455	460
Met	Arg	Arg	Ala	Ala	Lys	Thr	Ile	Asn	Phe	Gly	Val	Leu	Tyr	Gly	Met	465	470	475
Ser	Ala	His	Arg	Leu	Ser	Gln	Glu	Leu	Ala	Ile	Pro	Tyr	Glu	Glu	Ala	485	490	495
Gln	Ala	Phe	Ile	Glu	Arg	Tyr	Phe	Gln	Ser	Phe	Pro	Lys	Val	Arg	Ala	500	505	510
Trp	Ile	Glu	Lys	Thr	Leu	Glu	Glu	Gly	Arg	Arg	Arg	Gly	Tyr	Val	Glu	515	520	525
Thr	Leu	Phe	Gly	Arg	Arg	Arg	Tyr	Val	Pro	Asp	Leu	Glu	Ala	Arg	Val	530	535	540
Lys	Ser	Val	Arg	Glu	Ala	Ala	Glu	Arg	Met	Ala	Phe	Asn	Met	Pro	Val	545	550	555

Gln	Gly	Thr	Ala	Ala	Asp	Leu	Met	Lys	Leu	Ala	Met	Val	Lys	Leu	Phe
				565					570					575	
Pro	Arg	Leu	Glu	Glu	Met	Gly	Ala	Arg	Met	Leu	Leu	Gln	Val	His	Asp
			580					585					590		
Glu	Leu	Val	Leu	Glu	Ala	Pro	Lys	Glu	Arg	Ala	Glu	Ala	Val	Ala	Arg
		595					600					605			
Leu	Ala	Lys	Glu	Val	Met	Glu	Gly	Val	Tyr	Pro	Leu	Ala	Val	Pro	Leu
	610					615					620				
Glu	Val	Glu	Val	Gly	Ile	Gly	Glu	Asp	Trp	Leu	Ser	Ala	Lys	Glu	Gly
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Ile	Asp	Gly	Arg	Gly	Gly	Gly	Gly	His	His	His	His	His	His		
				645					650						

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 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:primer L71F

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<210> 14  
 <211> 20  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> Description of Artificial Sequence:primer L71R

<400> 14  
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<210> 15  
 <211> 22  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> Description of Artificial Sequence:primer L18015F

<400> 15  
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<210> 16  
 <211> 25  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> Description of Artificial Sequence:primer L23474R

<400> 16  
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<210> 17  
 <211> 22  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Description of Artificial Sequence:primer L18015F  
  
 <400> 17  
 tgacggagga taacgccagc ag 22

<210> 18  
 <211> 22  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Description of Artificial Sequence:primer L29930R  
  
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<210> 19  
 <211> 20  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Description of Artificial Sequence:primer L30350F  
  
 <400> 19  
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<210> 20  
 <211> 22  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
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 <400> 20  
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<210> 21  
 <211> 23  
 <212> DNA  
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 <220>  
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 <400> 21  
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<210> 22  
<211> 23  
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<220>  
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<400> 22  
cagcgggtgct gactgaatca tgg 23

<210> 23  
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<220>  
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<210> 24  
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<220>  
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<210> 25  
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<400> 25  
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<210> 26  
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<210> 27  
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beta-globin primer Bglbn536R

<400> 28  
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<210> 29  
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beta-globin primer Bglbn1408R

<400> 29  
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<210> 30  
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tag

<400> 30  
His His His His His His  
1 5

<210> 31  
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<212> PRT  
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<220>  
<223> Description of Artificial Sequence:anti-DYKDDDDK  
epitope tag

<400> 31  
Asp Tyr Lys Asp Asp Asp Asp Lys  
1 5

<210> 32  
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<220>  
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<400> 32  
Gly Gly Val Thr  
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<210> 33  
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<400> 33  
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<210> 34  
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<212> PRT  
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<220>  
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peptide

<400> 34  
Asn Ser Lys Lys Lys Lys Lys Lys Lys Arg Lys Lys Arg Lys Lys Lys  
1 5 10 15  
Gly Gly Gly Val Thr  
20